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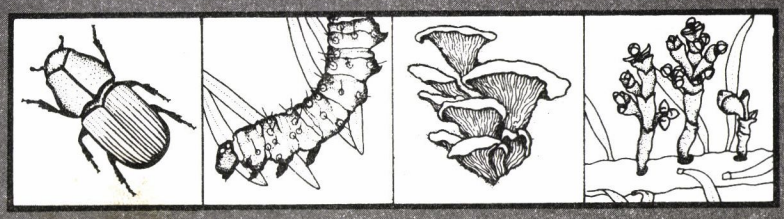

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CONTAINERIZED WESTERN WHITE PINE SEEDLING MORTALITY AT THE BONNERS FERRY RANGER DISTRICT, IDAHO PANHANDLE NATIONAL FORESTS

by

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ABSTRACT

Mortality of containerized western white pine seedlings outplanted on the Bonners Ferry Ranger District, Idaho Panhandle National Forests, was probably due to extensive root infection by Fusarium oxysporum. Diseased seedlings had chlorotic foliage, needle tip dieback, and severe twisting of the needles that indicated wilting. Most root tips of diseased seedlings were necrotic and colonized with F. oxysporum and Cylindrocarpon sp. First-year outplanting survival of seedlings varied among the different planting sites. Recommendations to reduce future losses are discussed.

INTRODUCTION

Western white pine (Pinus monticola Dougl.) seedlings from blister rust (Cronartium ribicola Fisch.) resistant seed sources are often grown in containers at the USDA Forest Service Nursery, Coeur d'Alene, Idaho, for outplanting on sites of relatively high blister rust hazard in both northern Idaho and northwestern Montana. Several thousand containerized white pine seedlings (seedlot 3378) were delivered to the Bonners Ferry Ranger District, Idaho Panhandle National Forests, in February 1984, for storage and outplanting in the spring of 1984. Seedlings were stored in a new enclosed facility at the Ranger Station at about 0-2°C (relative humidity 95-100 percent). Examination of the stored seedlings in March 1984 revealed that many seedlings were infected with the grey mold fungus, Botrytis cinerea Pers. ex Fr. However, mortality from Botrytis was only sporadic. To reduce further losses from this disease, seedlings were stored at lower temperatures (-1 to 0°C) until shipment for outplanting.

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Initial outplanting began in mid-April; seedlings with severe Botrytis infection were discarded, but those with light or moderate infection were usually planted. Several of the planting sites were examined for seedling survival about 3 weeks after planting. Most of the examined seedlings were dead or dying. Sample seedlings were sent to the Cooperative Forestry and Pest Management laboratory in Missoula for diagnosis of causes of death. Planting of seedlings from the affected seedlot continued on the District through June. This report summarizes findings regarding possible causes of poor seedling survival after outplanting and recommendations for improvement in the future.

MATERIALS AND METHODS

Isolations were made from the roots of several seedlings with different levels of foliar wilt symptoms; i.e., from slight foliar chlorosis and needle tip dieback to extensive foliar necrosis and needle twisting. Generally, a minimum of 15 fine root tips per seedling were placed on Komada's selective medium for Fusarium (Komada 1975) and incubated under cool fluorescent light at about 22°C for 5-7 days. Species of Fusarium were transferred to potato dextrose agar (PDA) for determination of growth characteristics (Booth 1971; Gerlach and Nirenberg 1982) and carnation leaf agar (CLA) for sporodochial production and sporulation (Fisher et al. 1982). Occurrence of fungi from four different genera (Fusarium, Cylindrocarpon, Trichoderma, and Penicillium) were tallied for each sampled seedling.

Seedling survival over a 4-month period was evaluated for one tray of containerized seedlings with 200 cells (198 live trees). Seedlings were kept within a growth chamber at 22-24°C under a 12-hour diurnal cycle of cool, fluorescent light and watered at least 3 days a week to prevent desiccation. Seedlings with severe wilt symptoms (chlorotic foliage, needle tip dieback, and needle twisting) were considered dead and isolations were made from the root tips of these seedlings on Komada's medium. Occurrence of Fusarium and Cylindrocarpon was tallied for each wilted seedling.

First-year outplanting survival was evaluated by District personnel on six sites planted with western white pine. Average survival (trees/acre) was tallied. Other factors such as site aspect, soil and air temperature, and relative humidity at the time of planting were compared with survival to evaluate possible relationships.

RESULTS

Relationships between intensity of foliar wilt symptoms and occurrence of selected fungi on the root tips of seedlings are outlined in table 1. Fusarium oxysporum Schlect. was isolated from seven of the nine seedlings analyzed (see Appendix for detailed description of this fungus). The two seedlings which did not yield F. oxysporum were infected with Cylindrocarpon sp. and extensively colonized with Trichoderma. The greatest percentage of root tips containing both F. oxysporum and Cylindrocarpon were from seedlings with moderate wilt symptoms. Although seedlings with light wilt symptoms contained some Fusarium, these seedlings often had a greater percentage of Trichoderma on their roots,

Table 1.—Relationships between intensity of foliage wilt symptoms and recovery of selected fungi from the root tips of containerized western white pine seedlings, Bonners Ferry Ranger District, Idaho Panhandle National Forests.

Seedling number	Foliar symptoms[1]	Percent Root Tip Infection				Other (unidentified)
		Fusarium	Cylindrocarpon	Trichoderma	Penicillium	
1	Extensive	10	7	77	7	3
2	Moderate	30	27	67	30	10
3	Extensive	30	3	43	17	17
4	Light	7	0	70	37	3
5	Moderate	53	0	80	7	6
6	Extensive	0	10	77	57	3
7	Light	0	20	100	7	0
8	Moderate	37	27	93	30	0
9	Light	23	3	87	23	7
-	Soil mix	0	0	100	7	0
Averages	Extensive	13.3	6.7	65.7	27.0	7.7
	Moderate	40.0	18.0	80.0	22.3	5.3
	Light	10.0	7.7	85.7	22.3	3.3

[1]Symptoms at time of isolate: Extensive = most foliage was necrotic with pronounced twisting of needles; Moderate = needle tips were necrotic, base of needles were green with moderate twisting of needles; Light = low level of foliage chlorosis, a few isolated needles were necrotic with little needle twisting evident.

Trichoderma was also commonly isolated from the soil mix samples; No Fusarium or Cylindrocarpon were recovered from these mix samples. Penicillium spp. were recovered with about equal frequency from all categories of wilted seedlings.

Approximately 27 percent of the seedlings evaluated for survival in a growth chamber died within four months (Table 2). More than 70 percent of the seedlings that died yielded F. oxysporum on their roots; almost a quarter of the seedlings also had Cylindrocarpon on some of their roots.

First-year outplanting survival of containerized western white pine seedlings was erratic among the six sites sampled (Table 3). Survival varied about 16 to 100 percent. Relationships with site aspect, soil and air temperature, and relative humidity were inconsistent. For example, on three sites with a southwestern aspect, survival was 16, 40, and 99 percent. Poor survival may have been due to transpiration stress from high air temperatures and low soil temperatures at the time of planting. Other factors, such as high levels of disease on roots, may have been partially responsible for poor survival.

Table 2.—Four-month survival of containerized western white pine seedlings[1] from the Bonners Ferry Ranger District, Idaho Panhandle National Forests.

Date	No. live seedlings	Percent survival	Percent sampled seedlings with *Fusarium* spp.	Percent sampled root tips with *Fusarium* spp.[2]	Percent dead seedlings with *Cylindrocarpon* spp.	Percent sampled root tips with *Cylindrocarpon* spp.[2]
5/18[3]	198	100.0	-	-	-	-
6/11	172	86.9	73.1	67.2	38.5	54.5
6/29	169	85.4	33.3	93.8	33.3	40.0
7/16	164	82.8	40.0	66.7	0	-
8/15	156	78.8	75.0	36.7	0	-
9/10	144	72.7	83.3	60.0	8.3	100.0
Totals for test	144	72.7	70.4	61.8	22.2	56.3

[1]Seedlings grown at 22-24°C under 12-hour cycles of cool, fluorescent light and watered at least three times a week.

[2]Includes only those seedlings from which *Fusarium* or *Cylindrocarpon* spp. were isolated.

[3]Start of the survival test.

Table 3.—First-year outplanting survival of containerized western white pine seedlings[1] on the Bonners Ferry Ranger District, Idaho Panhandle National Forests.

Unit number	Acres planted	Lot number	Aspect	Trees per acre			Temperature °C			Relative Humidity (%)
				Planted	Survived	Survival	Planted	Soil	Air	
703-2-26	48	3378	SW	208	34	16.3	6/13-18	9	16	60
703-2-29	32	3378	SW	366	146	39.9	6/12-13	6	17	50
720-3-42	24	3378	NE	175	175	100.0	6/20-27	11	29	31
703-3-103	8	3378	NW	775	442	57.0	6/11	8	17	40
703-6-34	20	3378	SW	333	330	99.1	6/11	8	18	29
703-6-36	24	3378	W	366	115	31.4	6/14-15	9	11	50
Average	-	-	-	370.5	207.0	55.9	-	8.5	16.3	43.3

[1]Survival at the end of the first growing season - counts taken in November.

DISCUSSION

This evaluation indicates that root disease caused by F. oxysporum was probably the major cause of mortality of containerized western white pine seedlings that occurred before or shortly after outplanting during the spring of 1984. Cylindrocarpon sp. also seemed to be associated with some of the seedling mortality. Although Cylindrocarpon spp. have previously been detected on diseased conifer seedlings (James and Gilligan 1985), their role in initiating disease is unclear.

Fusarium oxysporum is commonly isolated from the roots of diseased containerized seedlings (James 1984b; James and Gilligan 1985); intensity of damage may be related to level of seed infection which often varies among seedlots (James 1985). Apparently, infection by F. oxysporum can occur shortly after seedling emergence (Bloomberg 1966). However, disease symptoms may not appear until seedlings are much older, particularly during the last weeks of production or during storage and outplanting. It is suspected that root systems of white pine seedlings that died shortly after outplanting were heavily infected with F. oxysporum. They were probably not able to withstand periods of stress before and during outplanting. Even though F. oxysporum may die out when infected seedlings are outplanted in forest soils (Smith 1967), level of root infection was probably too severe for seedling recovery. Most of the killed seedlings were likely dead or nearly so when they were outplanted. Light or moderate wilt symptoms may not have been detected by planting crews and these seedlings were probably extensively infected by F. oxysporum.

Presence of B. cinerea on the foliage of containerized seedlings was quite common; infection probably occurred while the seedlings were still at the nursery. Extensive infection by B. cinerea may have reduced seedling vigor to the point where they were more susceptible to F. oxysporum. However, predispositional relationships between these two pathogens have not been established experimentally.

This evaluation indicated that Trichoderma spp. were generally more common on roots of seedlings with moderate or light wilt symptoms. Occurrence of Trichoderma on seedling roots may indicate exclusion or replacement of potential pathogens such as Fusarium and Cylindrocarpon.

Initial sources of F. oxysporum inoculum on the containerized pine seedlings are unknown. Although this fungus is commonly seedborne (James 1985), natural seed infection on seed orchard trees would be expected to be low. However, Fusarium infection could have been significant if cones contacted the soil for prolonged periods during collection and storage. Secondary spread of the pathogen within greenhouses from a few infected seed is likely, although disease symptoms may not have been evident. Another possible source of inoculum is contaminated peat-vermiculite soil mixes (James 1984a). However, assays indicated that pathogens were not found in sampled mixes.

It is recommended that seed from this white pine seedlot be screened for background levels of Fusarium contamination to determine if seed treatments with fungicides or sterilants are warranted to reduce future disease. An analysis of levels of root infection by Fusarium at different stages of seedling production and storage would provide valuable information on disease progression. Work to clarify roles of difference procedures that stress seedlings on level of root infection and subsequent outplanting survivability would also be useful.

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APPENDIX

Description of the Fusarium oxysporum isolate consistently obtained from wilted containerized western white pine seedlings:

Fusarium oxysporum Schlect. (Isolate 84-71)

- colonies fast growing, reaching 7.5 cm in diameter in 7 days at 22°C on PDA.
- colonies generally floccose, cream to flesh colored.
- colonies with slight violet to rose pigmentation, especially pronounced on agar surface.
- abundant microconidia produced, borne on short, mostly unbranched phialidic conidiophores, ellipsoid to oval, and measure 7-10 x 2.5-3.0 µ.
- macroconidia fusiform, moderately falcate, pointed at both ends, basal cells pedicellate, mostly 3 septate, and measure 30-45 x 3.0-4.0 µ.
- chlamydospores generally abundant, terminal or intercalary, globose to subglobose, and measure 7-11 µ in diameter.
- sporodochia containing mostly macroconidia formed on CLA with slight orange pigmentation.